# NEUROTRANSMITTERS

These chemicals released from nerve-fiber endings are the messengers
by means of which nerve cells communicate. Neurotransmitters mediate
functions ranging from muscle contraction to the control of behavior

by Julius Axelrod

In 1901 the noted English physiologist J. N. Langley observed that the injection of an extract of the adrenal gland into an animal stimulated tissues innervated by the sympathetic nerves: the nerves of the autonomic nervous system that increase the heart rate, raise the blood pressure and cause smooth muscles to contract. Just three years before that John J. Abel of Johns Hopkins University had isolated the hormone adrenaline from the adrenal gland, and so Langley's observation prompted T. R. Elliott, his student at the University of Cambridge, to inject adrenaline into experimental animals. Elliott saw that the hormone, like the crude extract, produced a response in a number of organs that was similar to the response evoked by the electrical stimulation of sympathetic nerves. He thereupon made the brilliant and germinal suggestion that adrenaline might be released from sympathetic nerves and then cause a response in muscle cells with which the nerves form junctions. Elliott thus first enunciated the concept of neural communication by means of chemical transmitters. A neurotransmitter is a chemical that is discharged from a nerve-fiber ending. It reaches and is recognized by a receptor on the surface of a postsynaptic nerve cell or other excitable postjunctional cell and either stimulates or inhibits the second cell. Today it is clear that many different neurotransmitters influence a variety of tissues and physiological processes. Neurotransmitters make the heart beat faster or slower and make muscles contract or relax. They cause glands to synthesize hormone-producing enzymes or to secrete hormones. And they are the agents through which the brain regulates movement and changes mood and behavior.

Elliott's concept of chemical neurotransmission was accepted slowly. Lang-ley, who disliked theories of any kind, discouraged further speculation by Elliott until more facts were available. That took time. The first definite evidence for neurochemical transmission was obtained in 1921 by Otto Loewi, who was then working at the University of Graz in Austria, through an elegant and crucial experiment. Loewi put the heart of a frog in a bath in which the heart could be kept beating. The fluid bathing the heart was allowed to perfuse a second heart. When Loewi stimulated the first heart's vagus nerve (a nerve of the parasympathetic system that reduces the heart rate), the beat of the second heart was slowed, showing that some substance was liberated from the stimulated vagus nerve, was transported by the fluid and influenced the perfused heart. The substance was later identified by Sir Henry Dale as acetylcholine, one of the first neurotransmitters to be recognized. In a similar experiment the stimulation of the accelerans nerve (the sympathetic nerve that increases the heart rate) of a frog heart speeded up the beat of an unstimulated perfused heart. In 1946 the Swedish physiologist Ulf von Euler isolated the neurotransmitter of the sympathetic system and identified it as noradrenaline.

## The Transmitters

To be classed as a neurotransmitter a chemical should fulfill a certain set of criteria. Nerves should have the enzymes required to produce the chemical; when nerves are stimulated, they should liberate the chemical, which should then react with a specific receptor on the postjunctional cell and produce a biological response; mechanisms should be available to terminate the actions of the chemical rapidly. On the basis of these criteria two compounds are now estab-lished as neurotransmitters: acetylcholine and noradrenaline. Nerves that contain them are respectively called cholinergic and noradrenergic nerves. There are a number of other nerve chemicals that meet many of the listed criteria but have not yet been shown to meet them all. These "putative" transmitters are dopamine, adrenaline, serotonin, octopamine, histamine, gamma aminobutyric acid, glutamic acid, aspartic acid and glycine.

This article will deal mainly with one class of neurotransmitters, the catecholamines, since more is known about these compounds than about some other transmitters and since many of the principles governing their disposition appear to govern those of transmitters in general. The catecholamines include noradrenaline (also known as norepinephrine), dopamine and adrenaline (or epinephrine). They have in common a chemical structure that consists of a benzene ring on which there are two adjacent hydroxyl groups and an ethylamine side chain. Noradrenaline is present in peripheral nerves, the brain and the spinal cord and in the medulla, or inner core, of the adrenal gland. In peripheral tissues and in the brain noradrenaline acts as a neurotransmitter, that is, it exerts most of its effect locally on postjunctional cells. In the adrenal medulla it functions as a hormone, that is, it is released into the bloodstream and acts on distant target organs. Dopamine, once thought to be simply an intermediate in the synthesis of noradrenaline and adrenaline, is also a neurotransmitter in its own right in the brain, where it functions in nerves that influence movement and behavior. The third catecholamine, adrenaline, is largely concentrated in the adrenal medulla. It is discharged into the bloodstream in fear, anger or other stress and acts as a hormone on a number of organs, includ-

ing the heart, the liver and the intestines. Just in the past year it has developed that adrenaline is probably also a neurotransmitter, since it is found in nerves in the brain.

Techniques developed a decade ago in Sweden made it possible to visualize catecholamines in neurons directly, by the fluorescent glow they emit after treatment with formaldehyde vapor. Fluorescence photomicrography, electron microscopy and radioautography have revealed the structure and functioning of the sympathetic nerve cell in great detail [see illustration below]. The neuron has a cell body and a long axon, or main fiber, that branches into a large number of terminals. Each nerve ending is studded with varicosities, or swellings, that look like beads on a string, so that a single sympathetic neuron can innervate thousands of other cells: "effector" cells.

In 1960 Georg Hertting, Gordon Whitby and I were able to show that radioactive noradrenaline (noradrenaline in which tritium, the radioactive isotope of hydrogen, has been substituted for some of the hydrogen atoms) is taken up selectively and retained in sympathetic nerves. In my laboratory at the National Institute of Mental Health we went on to find out where the neurotransmitter is stored within the nerve cell. Electron

micrographs of sympathetic-neuron varicosities reveal large numbers of vesicles with dark, granular cores. When photographic film was exposed to tissues from rats injected with labeled noradrenaline, the silver grains developed by the radiation from the radioactive hydrogen atoms were strikingly localized over the granulated vesicles [see illustrations on opposite page]. This indicated that it is in those vesicles that noradrenaline is stored within the nerve.

## Synthesis and Release

The process leading to the synthesis of catecholamine transmitters begins in the cell body, which has the machinery for making the four enzymes needed for their formation: tyrosine hydroxylase, dopa decarboxylase, dopamine beta-hydroxylase (DBH) and phenylethanolamine N-methyltransferase (PNMT). The enzymes synthesized in the cell body are carried down the axon by a natural flowing process to the nerve endings, where the synthesis of the catecholamines is achieved.

The discharge of neurotransmitter from the nerve endings caps a complex series of events. When a nerve is stimulated, its membrane is depolarized, with sodium moving into the nerve as potassium comes out; the nerve signal propa-

gates as a wave of depolarization that moves along the nerve axon to the endings. As Bernhard Katz of University College London first showed for acetylcholine, the depolarization causes a quantum—a packet or spurt, as it were—of the transmitter to be discharged from the nerve ending into the synaptic cleft.

Biochemical evidence recently obtained in our laboratory and others shows that noradrenaline is released from nerves in much the same way. The vesicles in the endings contain not only noradrenaline but also the enzyme, DBH, that converts dopamine into noradrenaline. When the sympathetic nerve is stimulated electrically, noradrenaline and the enzyme are released in about the same proportions in which they are present in the vesicles. The only way that could happen would be through the fusion of the vesicle with the outer membrane of the nerve, followed by the formation of an opening large enough to allow molecules of noradrenaline to be extruded along with the much larger molecules of the enzyme. Such a release mechanism is called exocytosis. The detailed events whereby the vesicle fuses with the neural membrane and makes an opening to discharge its soluble contents are uncertain, as is the subsequent fate of the vesicle. We do know that certain conditions prevent the release of noradrenaline and DBH. One is the presence of vinblastin, a compound that breaks down the protein structures in nerve cells called neurotubules. Another is the presence of cytocholasin-beta, a substance that disrupts the function of the contractile filament system in cells. A third is the absence of calcium. These findings suggest that the long, tubelike protein structures may orient the vesicles to a site on the neuronal membrane from which the release occurs. It is well known that microfilaments in cells other than nerves, such as muscle cells, can be activated by calcium so that they contract. It is therefore possible that depolarization causes calcium to activate a contractile filament on the neural membrane, which thereupon contracts to make an opening large enough so that the soluble contents of the vesicle can be discharged.

The observation that DBH is released from nerves suggested to Richard Weinshilboum, a research associate in my laboratory, that the enzyme might find its way into the bloodstream. We devised a sensitive assay for the enzyme and found it is indeed present in the blood, and we and others went on to measure the amount of the enzyme (which is found specifically in sympathetic nerves) in a
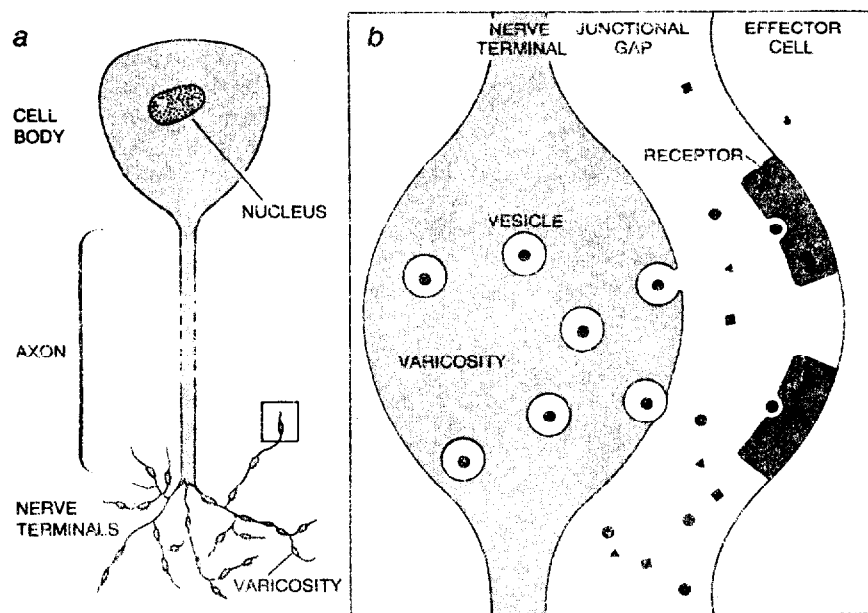


SYMPATHETIC NEURON, or nerve cell (a), consists of a cell body, a long axon and numerous nerve terminals studded with varicosities. Enzymes involved in the synthesis of the transmitters are made in the nucleus and transported down the axon to the varicosities, where the transmitters are manufactured and stored. A varicosity and its junction with another cell are enlarged (b). The transmitter (colored dots) is stored in vesicles. When the nerve is excited and depolarized, vesicles move to the cell membrane and fuse with it, releasing transmitter into the junctional gap. The transmitter reaches receptors on the effector cell that recognize only it, not any other chemicals (black shapes) that are present in the gap.
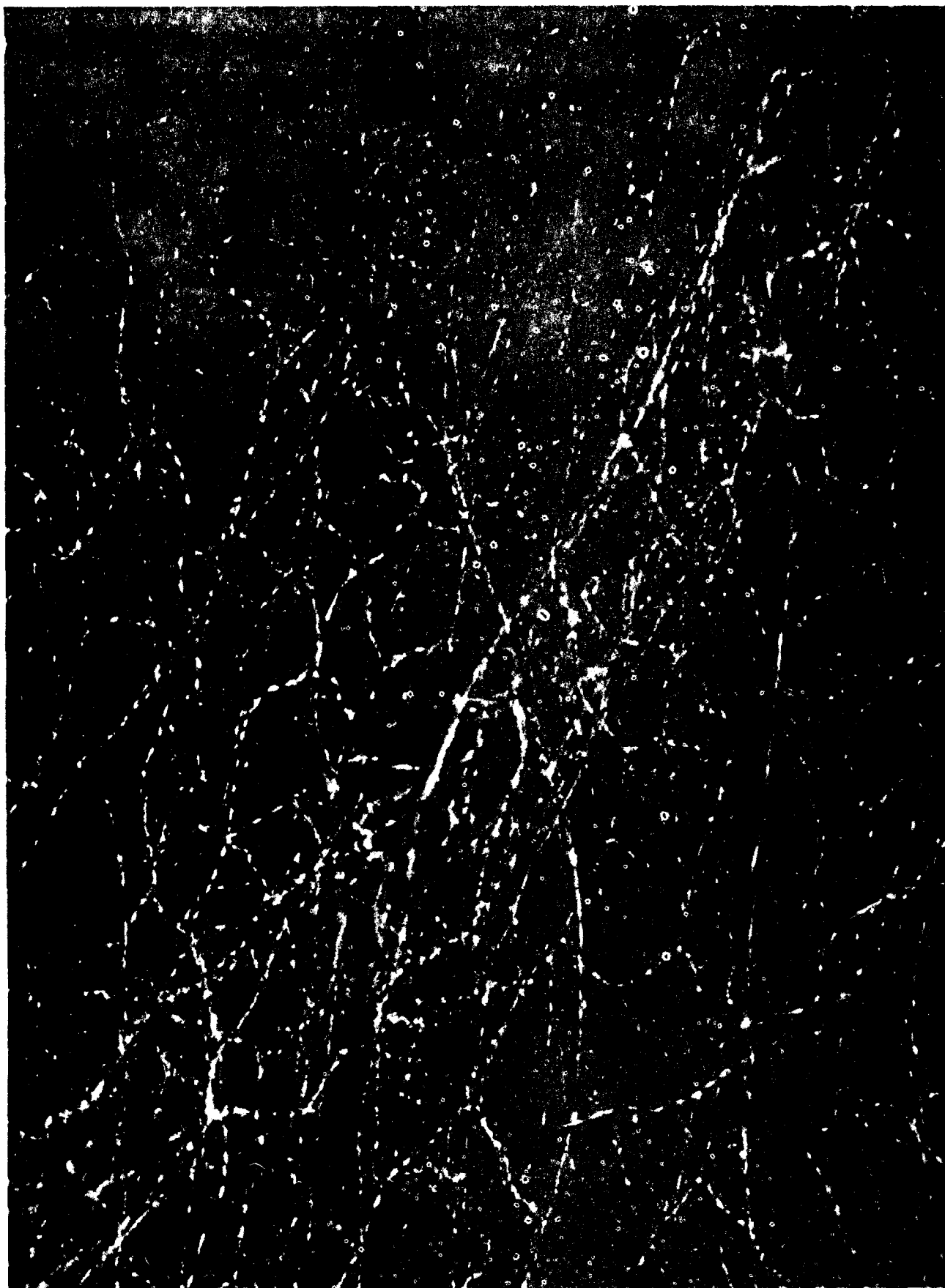
SYMPATHETIC-NERVE TERMINALS from the iris of a rat's eye emit a green glow after treatment with formaldehyde, showing that they contain noradrenaline, one of the neurotransmitters. The ter- minals, studded with varicosities where the noradrenaline is stored, are enlarged 2,400 diameters in this fluorescence micrograph made by David Jacobowitz of the National Institute of Mental Health.
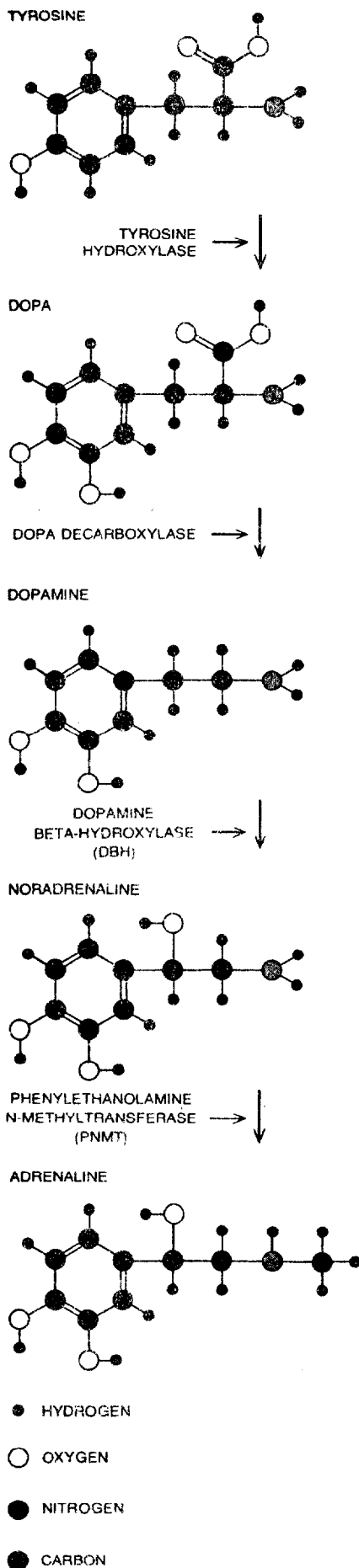
VARICOSITIES on noradrenaline-containing nerve endings from a rat pineal gland are enlarged 90,000 diameters in an electron mi- crograph made by Floyd Bloom of the National Institute of Mental Health. The varicosities contain vesicles, many with dense cores.



RADIOACTIVE NORADRENALINE is shown by radioautogra- phy to be localized in the vesicles. A pineal-tissue sample was tak- en from rats injected with labeled noradrenaline. The radioactivity developed silver grains (black blots) in a photographic film laid on the sample. The developed grains were strikingly localized over the vesicles, which were thus identified as sites of transmitter storage.

TYROSINE

TYROSINE
HYDROXYLASE →

DOPA

DOPA DECARBOXYLASE →

DOPAMINE

DOPAMINE
BETA-HYDROXYLASE →
(DBH)

NORADRENALINE

PHENYLETHANOLAMINE
N-METHYLTRANSFERASE →
(PNMT)

ADRENALINE

● HYDROGEN
○ OXYGEN
● NITROGEN
● CARBON

variety of disease states. It is low in the hereditary disorder of the autonomic system called familial dysautonomia and in Down's syndrome (mongolism), and it is high in torsion dystonia (a neurological disease involving muscle spasticity), in neuroblastoma (a cancer of nervous tissue) and in certain forms of hypertension. The findings suggest that in each of these diseases there are abnormalities in the functioning of the sympathetic nervous system.

## Action and Inactivation

Once the neurotransmitter is liberated it diffuses across the cleft between the nerve terminal and adjacent cells. The capacity of a neighboring effector cell to respond to the transmitter then depends on the ability of a receptor on the postjunctional cell's surface to selectively recognize and combine with the neurotransmitter. When the receptor and transmitter interact, a series of events is triggered that causes the effector cell to carry out its special function. Some of these responses occur rapidly (in a fraction of a second), as in the propagation of nerve transmission across a synapse; others occur slowly, in minutes or sometimes hours, as in the synthesis of intracellular enzymes. There are two receptors that recognize noradrenaline, alpha and beta adrenergic receptors, and there is one for dopamine. These receptors can be distinguished from one another by the specific response each elicits and by the ability of specific drugs to block those responses.
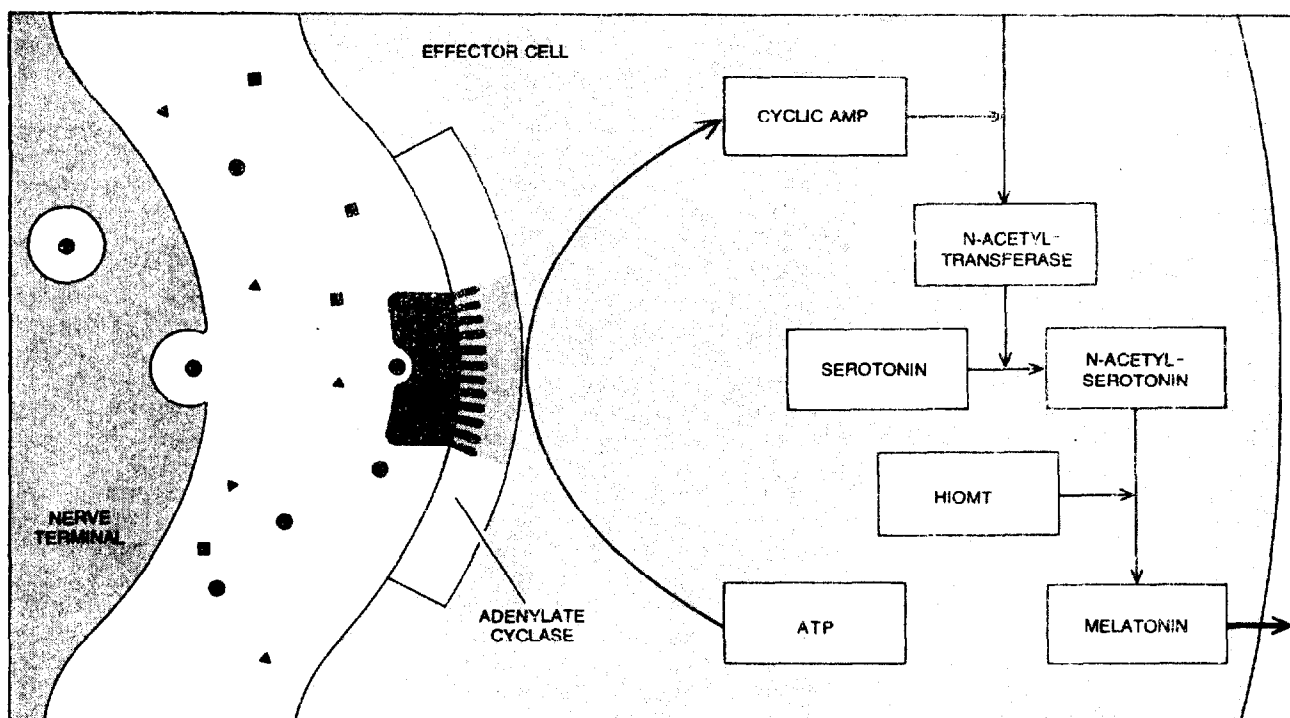
The beta adrenergic receptors turn on an effector cell, and they do so by means of adenosine 3'5' monophosphate, or cyclic AMP, the universal "second messenger" that mediates between hormones and many cellular activities elicited by those hormones [see "Cyclic AMP," by Ira Pastan; SCIENTIFIC AMERICAN, August, 1972]. Investigators have traced several of the steps in the activation of the receptor by noradrenaline by studying the interaction of noradrenaline and fat cells or cells of the liver or of the

PRODUCTION of catecholamine transmitters is accomplished by four enzymes (color) in sympathetic-nerve terminals and in the adrenal gland. Tyrosine, an amino acid, is transformed into the intermediate dopa. Removal of a carboxyl (COOH) group forms dopamine, which is itself a transmitter and is also the precursor of the transmitter noradrenaline. In the adrenal gland the process continues with the addition of a methyl ($CH_3$) group to form adrenaline.

pineal gland. We have found the pineal, which makes a hormone called melatonin that inhibits the activity of sex glands, particularly suitable because it is heavily supplied with nerves containing noradrenaline [see "The Pineal Gland," by Richard J. Wurtman and Julius Axelrod; SCIENTIFIC AMERICAN, July, 1965]. Melatonin is synthesized in a number of steps, one of which, the conversion of serotonin to N-acetylserotonin, is catalyzed by the enzyme serotonin N-acetyltransferase. It is that enzyme's synthesis that is controlled by the beta adrenergic receptor. When noradrenaline is released from a nerve innervating the pineal, it interacts with beta adrenergic receptors on the outside of the membrane of a pineal cell. Once a receptor is occupied by noradrenaline, the enzyme adenylate cyclase, on the inner surface of the cell membrane, is activated. The adenylate cyclase then converts the cellular energy carrier ATP to cyclic AMP, which in turn stimulates the synthesis of serotonin N-acetyltransferase [see illustration on opposite page]. This complex series of events can be turned off by propranolol, a drug that prevents the noradrenaline from combining with the beta adrenergic receptor.

The adenylate cyclase system is involved in scores of biological actions. The ability of the pineal cell to carry out its special function, the manufacture of melatonin, by utilizing the almost universal adenylate-cyclase system depends on the presence of receptors on the cell surface that can specifically recognize noradrenaline and of the enzyme hydroxyindole-O-methyltransferase, uniquely present in the pineal cell, that can convert N-acetylserotonin to melatonin.

Once the neurotransmitter has interacted with the postjunctional cell, its actions must be rapidly terminated; otherwise it would exert its effects for too long and precise control would be lost. In the cholinergic nervous system the acetylcholine is rapidly inactivated by the enzyme acetylcholinesterase, which metabolizes the transmitter. In the past 10 years it has become clear that the inactivation of neurotransmitters through enzymatic transformation is the exception rather than the rule. Catecholamine neurotransmitters are metabolized by two enzymes, catechol-O-methyltransferase (COMT) and monoamine oxidase (MAO); the latter is a particularly important enzyme that removes the amino ($NH_2$) group of a wide variety of compounds, including serotonin, noradrenaline, dopamine and adrenaline. There are enzyme-inhibiting chemicals that

MODE OF ACTION of a transmitter is exemplified by the effect of noradrenaline on a pineal cell. Noradrenaline (*colored dots*) released from a nerve ending binds to a beta-adrenergic receptor on the pineal-cell surface. The receptor thereupon activates the enzyme adenylate cyclase on the inside of the pineal-cell membrane. The activated adenylate cyclase catalyzes the conversion of adenosine triphosphate (ATP) into cyclic adenosine monophosphate (AMP). The cyclic AMP stimulates synthesis of the enzyme N-acetyltransferase; the enzyme converts serotonin into N-acetylserotonin. This is transformed in turn by the pineal cell's specific enzyme, hydroxyindole-O-methyltransferase (HIOMT), to form melatonin, the pineal-gland hormone that acts on the sex glands.

can prevent COMT and MAO from carrying out their biochemical transformations; when such inhibitors were administered, the action of noradrenaline was found still to be rapidly terminated. There had to be a method of rapid inactivation other than enzymatic transformation.

In order to track down such a mechanism we injected a cat with radioactive noradrenaline. The labeled transmitter persisted in tissues that were rich in sympathetic nerves for many hours, long after its physiological actions were ended, indicating that radioactive noradrenaline was taken up in the sympathetic nerves and held there. My colleagues and I designed a simple experiment to prove this. Sympathetic nerves innervating the left salivary gland in rats were destroyed by removing the superior cervical ganglion on the left side of the neck; about seven days after this operation the noradrenaline nerves of the salivary gland on the right side were intact, whereas the nerves on the left side had completely disappeared. When radioactive noradrenaline was injected, the transmitter was found in the right salivary gland but not in the left one. We also found that in cats injected with radioactive noradrenaline the transmitter was re-

leased when the sympathetic nerves were stimulated electrically. The experiments clearly demonstrated that noradrenaline is taken up into, as well as released from, sympathetic nerves. As a result of these experiments we postulated that noradrenaline is rapidly inactivated through its recapture by the sympathetic nerves; once it is back in the nerves, of course, the neurotransmitter cannot exert its effect on postjunctional cells. Leslie L. Iversen of the University of Cambridge has since shown that this neuronal recapture by sympathetic nerves is highly selective for noradrenaline or compounds resembling it in chemical structure. Recent work indicates that uptake by nerves may be the most general mechanism for the inactivation of neurotransmitters.

## Regulation

Chemical transmitters in sympathetic nerves (and presumably in other nerves) are in a state of flux, continually being synthesized, released, metabolized and recaptured. The activity of nerves can also undergo marked fluctuation during periods of stress. In spite of all these dynamic changes the amount of catecholamines in tissues remains constant.

This is owing to a variety of adaptive mechanisms that alter the formation, release and response of catecholamines. There are fast regulatory changes that require only fractions of a second and slower changes that take place after minutes or even hours.

When sympathetic nerves are stimulated, the conversion of tyrosine to noradrenaline in them is rapidly increased. The increased nervous activity specifically affects tyrosine hydroxylase, the enzyme that converts tyrosine to dopa, because its activity is inhibited by noradrenaline and dopamine. Any increase in nerve-firing brought on by stress, cold and certain drugs lowers the level of catecholamines in the nerve terminals. This reduces the negative-feedback effect of noradrenaline and dopamine on tyrosine hydroxylase, so that more tyrosine is converted to dopa, which in turn is converted to make more catecholamines. Conversely, when nerve activity is decreased, the catecholamine level rises, slowing down the conversion of tyrosine to dopa by once again inhibiting the tyrosine hydroxylase.

Another rapid regulation is accomplished at the nerve terminal itself, where the alpha adrenergic receptors are situated. When the alpha receptors are

activated, they diminish the release of noradrenaline from nerve terminals into the synaptic cleft. When too much noradrenaline is released, it accumulates in the synaptic cleft; when the catecholamine level is high enough, it activates the alpha receptors on the presynaptic nerve terminals and shuts off further release of the neurotransmitter.

A slower regulatory process is brought on by prolonged firing of sympathetic nerves, which can step up the manufacture of the catecholamine-synthesizing enzymes tyrosine hydroxylase, DBH and (to a lesser extent) PNMT; the rise in the enzyme level enables the nerves to make more neurotransmitter. We discovered this phenomenon of increased enzyme synthesis when we gave animals reserpine, a versatile drug that lowers the blood pressure and incidentally increases sympathetic-nerve firing (which tends to raise the pressure) by a reflex action. The reserpine brought about a gradual increase in tyrosine hydroxylase and DBH in sympathetic nerves and the adrenal gland and of PNMT in the adrenal gland. Increases in these enzymes were also found in animals exposed to stress, cold, physical restraint, psychosocial stimulation or insulin injection. When the synthesis of proteins was prevented by drugs, on the other hand,

there was no elevation in enzyme activity after reserpine was given. This indicated that increased nerve activity stimulates the synthesis of new molecules of tyrosine hydroxylase, DBH and PNMT; with a greater need for neurotransmitters there is a compensatory increase in the synthesis of enzymes that catalyze the making of these transmitters.

In order to learn whether the command for increased synthesis of new tyrosine hydroxylase and DBH molecules can be transmitted from one nerve to another we cut the nerve innervating certain noradrenaline cell bodies—the superior cervical ganglia—on one side. When nerves were then stimulated reflexly by reserpine, there was an elevation of tyrosine hydroxylase and DBH levels in the innervated ganglia but not in the denervated ones. The experiment showed that one nerve can transmit information to another nerve (presumably by means of a chemical signal) that causes the postsynaptic nerve to make new enzyme molecules.

## Sensitivity

In 1855 the German physiologist J. L. Budge observed that when the nerves leading to a rabbit's right eye were destroyed, the pupil of that eye became

more dilated than the left pupil. The phenomenon was later explained by the American physiologist Walter B. Cannon, who postulated that as a result of denervation the effector cells somehow become more responsive. He called this effect the "law of denervation supersensitivity." Subsequent work showed that denervation supersensitivity is caused by two separate mechanisms, one presynaptic and the other postsynaptic. When nerves are destroyed, presynaptic inactivation by recapture is abolished, thereby leaving the neurotransmitters to react with the postsynaptic site longer.

Denervation also causes a profound change in the degree of activity of the postjunctional cell. Recent work with the pineal gland in our laboratory has suggested a hypothesis for supersensitivity, and also for subsensitivity, in postjunctional cells. As we have seen, noradrenaline stimulates the synthesis of the enzyme serotonin N-acetyltransferase through a beta adrenergic receptor in the postjunctional pineal cell. When the nerves to pineal cells are destroyed (or depleted of noradrenaline by the administration of reserpine), the pineal cells become 10 times as responsive to noradrenaline; that is, when the postjunctional cell is deprived of its neurotransmitter for a period of time, it takes just
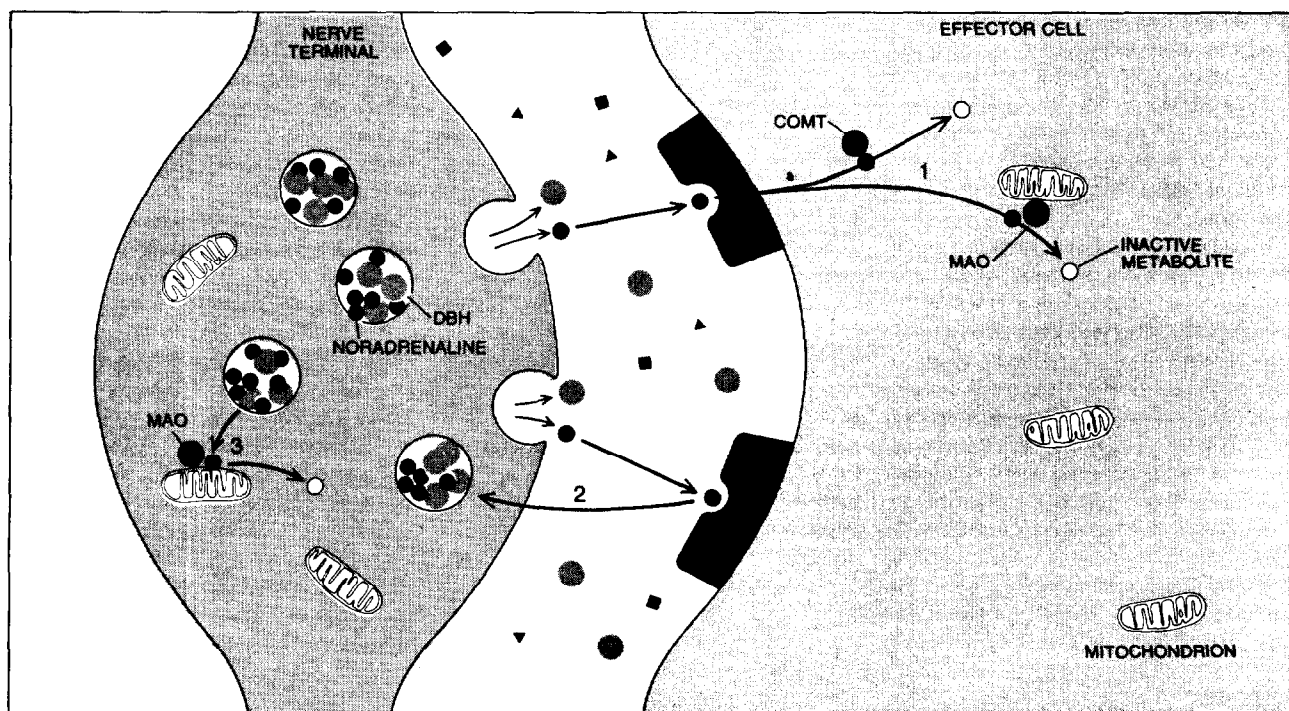


RELEASE AND INACTIVATION of the neurotransmitter noradrenaline is shown in more detail. The enzyme DBH is stored in the noradrenergic nerve terminals along with the transmitter and is released with it into the junctional gap. The noradrenaline binds to the receptors on the effector cell, eliciting that cell's response as shown in the illustration on the preceding page. Then the norad-

renaline's action is terminated either through metabolism (1) by the enzymes catechol-O-methyltransferase (COMT) and/or monoamine oxidase (MAO), or by recapture and storage (2) in the presynaptic sympathetic-nerve terminal; the latter is the more important process. MAO, stored in the membrane of mitochondria, also inactivates noradrenaline that leaks out of vesicles (3).

one packet of catecholamines to cause the same increase of N-acetyltransferase in the cell as 10 packets of transmitters would cause in a normally innervated cell. If, on the other hand, the pineal cell is exposed to an excessive amount of catecholamines for a period of time, it becomes less responsive: a larger amount of the transmitter is required to produce the same increase in N-acetyltransferase.

These experiments suggest that changes in the responsiveness of excitable cells are the result of an alteration in the "avidity" with which the receptor binds the neurotransmitters. If the receptor is exposed to small amounts of catecholamine for some time, it reacts with the neurotransmitter easily; if too many neurotransmitter molecules bombard the receptor, it becomes less responsive. Depending on the tissue, this change in sensitivity can come within hours or days, so that it is an effective adaptive mechanism for excitable cells. It is possible that the tolerance that is often developed to a drug taken in excess may reflect subsensitivity on the part of cells that respond to the drug.

## Role in the Brain

The brain has billions of nerves that talk to one another by means of neurotransmitters. Neurobiologists are just beginning to unravel the complex biochemistry and physiology of chemical transmission in the brain. Many different neurotransmitters function in brain neurons, but because there are more precise methods of measuring catecholamines and drugs are available that perturb their formation, storage, release and metabolism, we know more about brain catecholamines than about the other neurotransmitters. Fluorescence photomicrography and drugs that selectively destroy catecholamine-containing nerves have made it possible to locate the noradrenaline, dopamine and serotonin cell bodies and trace the pathways of their axons and nerve endings [see illustrations on page 71]. The cell bodies of the dopamine-containing nerves are in the area of the brain stem called the substantia nigra, whence the dopaminergic axons course through the brain stem; many of them terminating in the caudate nucleus. The dopamine-containing tracts in the caudate nucleus play an important role in the integration of movement.

The elucidation of the biochemistry and pharmacology of dopamine in the brain has led to the development of a powerful treatment of a crippling disease, Parkinsonism. The Swedish pharmacologist Arvid Carlsson noted in 1959
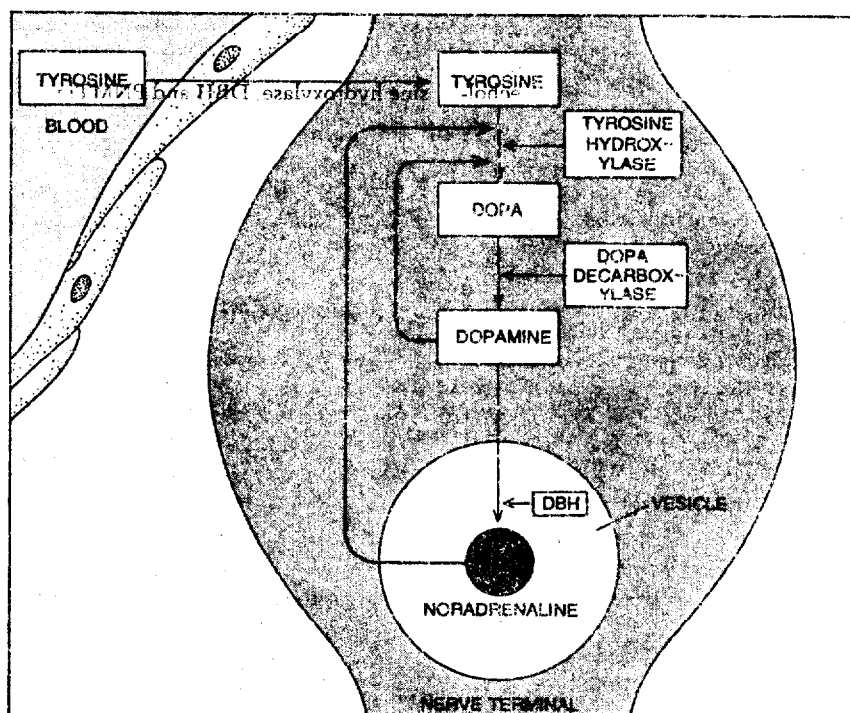


**RAPID REGULATION** of catecholamine synthesis is accomplished by a feedback mechanism: a buildup of dopamine and noradrenaline inhibits (*colored arrows*) the activity of tyrosine hydroxylase, which catalyzes the first step in the synthesis. An increase in nerve activity reduces the amount of dopamine and noradrenaline in the terminal, removing the inhibition; tyrosine hydroxylase activity increases and more transmitter is synthesized.

that when reserpine was given to rats, it sharply reduced the dopamine content of the caudate nucleus in the brain and also caused a Parkinson-like tremor. The administration of dopa, a dopamine precursor that can get from the blood into the brain more easily than dopamine, reversed the tremors. These findings prompted Oleh Hornykiewicz, who was then working at the University of Vienna, to measure the content of dopamine in the brain of patients who had died of Parkinson's disease. He found that there was virtually no dopamine in the caudate nucleus. The finding led directly to a major therapeutic advance by George C. Cotzias of the Brookhaven National Laboratory: when dopa, the dopamine precursor that can cross the blood-brain barrier, is administered, it makes up the dopamine deficiency and effectively relieves the symptoms of Parkinson's disease. This is a good example of how basic research can sometimes lead rapidly to a new treatment for a disease.

There are two main nerve tracts containing noradrenaline in the brain, the dorsal and ventral pathways. The cell bodies of the noradrenaline-containing tracts are found in the lower part of the brain in the area called the locus ceruleus, or "blue place." Noradrenaline-containing nerve tracts are highly branched

and reach many parts of the brain. Among the areas they innervate are the cerebellum and the cerebral cortex, which are concerned with the fine coordination of movement, alertness and emotion. Another part of the brain innervated by noradrenaline neurons is the hypothalamus, which controls many visceral functions of the body such as hunger, thirst, temperature regulation, blood pressure, reproduction and behavior. Manipulation of the noradrenaline levels in the brain can change many of the functions of the hypothalamus, particularly the "pleasure" centers. Noradrenaline tracts also appear to be involved in mood elevation and depression. Recently nerves containing adrenaline have also been observed in the brain stem. The next few years should show whether these adrenaline-containing tracts also control emotion, mood and behavior.

Drugs have been powerful tools for probing the action of neurotransmitters. As our knowledge concerning neurotransmitters has broadened, so has our understanding of the action of drugs on behavior and on the cardiovascular and motor systems; the two trends have interacted nicely. In the early 1950's pharmacologists recognized that the hallucinogenic agent lysergic acid diethylamine (LSD) not only resembles serotonin

in chemical structure but also counteracts some of its pharmacologic actions (by occupying sites intended for serotonin). Several workers therefore proposed that serotonin must have something to do with insanity. Other hallucinogenic agents such as mescaline and amphetamine, on the other hand, are related in structure to noradrenaline. In the mid-1950's clinical investigators were learning that chemicals such as chlorpromazine could mitigate psychotic behavior, and that monoamine oxidase inhibitors and imipramine and related drugs could relieve depression. At about the same time it was observed that reserpine, which was proving valuable not only for hypertension but also for schizophrenia, markedly reduced the levels of norad-renaline and serotonin in the brain. The observations combined to suggest that these drugs exerted their actions on the brain by interfering with neurotransmitters. When my colleagues and I found that radioactive noradrenaline can be taken up and released from nerves, we were in a good position to investigate how a drug influences the disposition of injected radioactive transmitters.

## Effect of Drugs

The first compound we examined was cocaine, a potent stimulant that can produce psychosis and that also intensifies the action of noradrenaline. When radioactive noradrenaline was injected into cats that had been given cocaine, the uptake of catecholamines by the sympathetic nerves was prevented, demonstrating that cocaine magnifies the effect of noradrenaline by preventing its capture and inactivation and leaving larger amounts of the catecholamine to react with the effector cell. Antidepressant drugs such as imipramine had the same effect: they blocked the uptake of noradrenaline into sympathetic nerves. By using radioactive noradrenaline we found that amphetamine, which is both a stimulant and a mind-altering drug, affects noradrenergic nerves in two ways: it blocks the uptake of noradrenaline and also promotes the release of the neurotransmitter from nerves.

Many drugs that are effective in the treatment of hypertension affect the
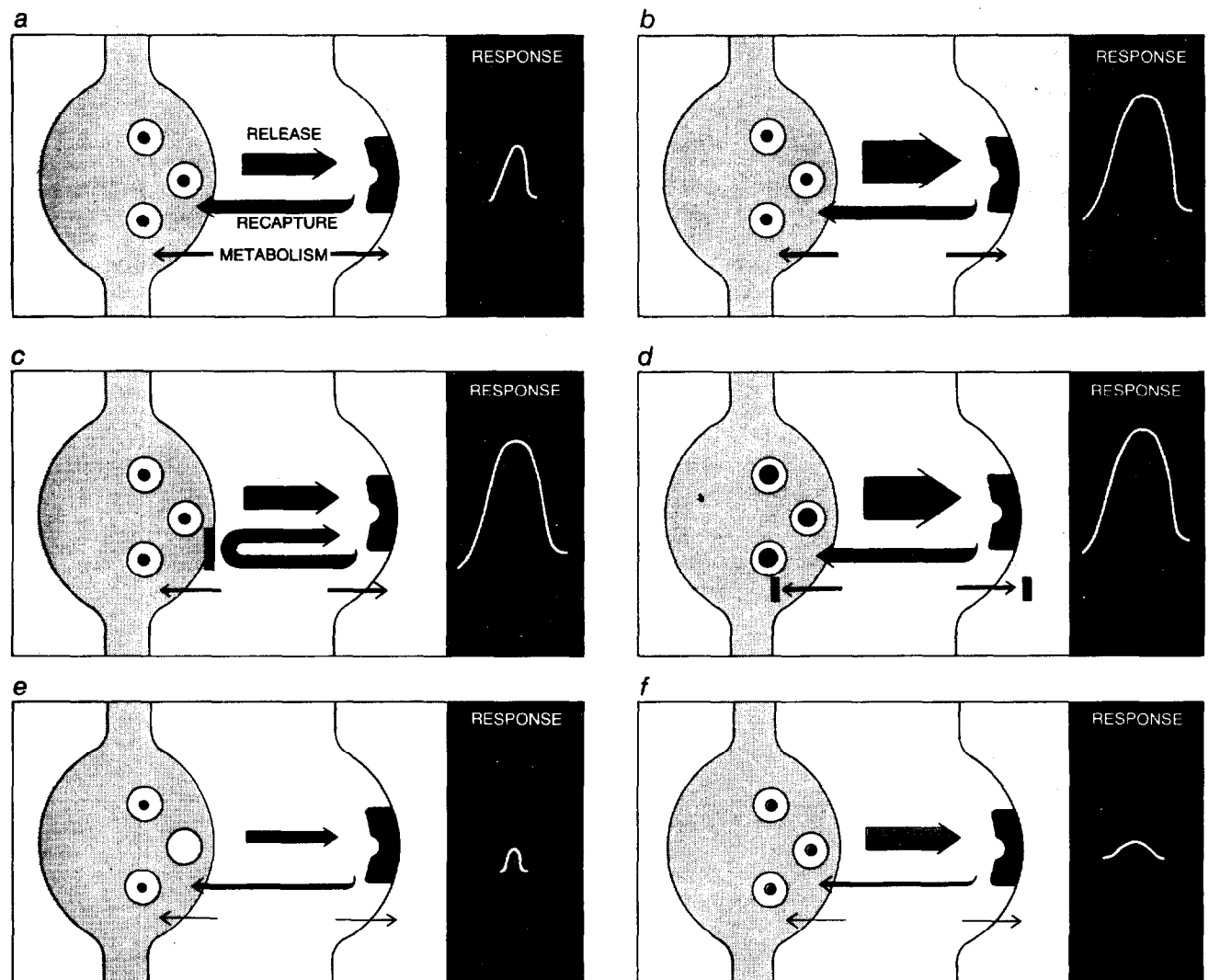
CERTAIN ADRENERGIC DRUGS increase or decrease the availability of noradrenaline at the adrenergic receptor. Normal release, recapture and metabolism (*colored arrows*) are illustrated, with a curve representing the normal response of a postjunctional cell (*a*). Antidepressant drugs enlarge that response in several ways, all of which increase the availability of noradrenaline at the synapse. Amphetamine does so by promoting the release of noradren-aline (*b*). Amphetamine and imipramine and related drugs block recapture (*c*); the monoamine-oxidase inhibitors interfere with inactivation through metabolism (*d*). Conversely, reserpine, which reduces blood pressure and may induce depression, reduces the response by depleting the noradrenaline in storage (*e*); alpha-methyldopa and other "false transmitters" are stored in the vesicles with noradrenaline and released with it, diluting its effect (*f*).

storage and release of noradrenergic transmitters. Reserpine and guanethidine reduce blood pressure by preventing the nerves that raise the pressure from storing noradrenaline. Antihypertensive drugs such as alpha methyl dopa, on the other hand, are transformed by enzymes in the nerve into substances that resemble the noradrenaline chemically. The "false transmitters" are stored and released along with natural neurotransmitters, diluting them and thus reducing their effect.

In the past 10 years many psychiatrists and pharmacologists have been struck by the fact that drugs that relieve mental depression also interfere with the uptake, storage, release or metabolism of noradrenaline. Whereas imipramine blocks the uptake of noradrenaline by nerves and amphetamine both releases noradrenaline and blocks its uptake, monoamine oxidase inhibitors, as their name implies, prevent the metabolism of the catecholamine. In other words, all these antidepressants produce similar results by different mechanisms: they increase the amount of catecholamine in the synaptic cleft, with the result that more transmitter is available to stimulate the receptor. Conversely, reserpine, a compound that decreases the amount of the chemical transmitters, sometimes produces depression. These considerations led to the proposal of a catecholamine hypothesis of depressive states, which holds that mental depression is associated with the decreased availability of brain catecholamine and is relieved by drugs that increase the amount of these transmitters at the adrenergic receptor. Although the hypothesis is not yet entirely substantiated, it has provided a valuable framework within which new approaches to understanding depression can be sought.

The introduction in the 1950's of antipsychotic drugs such as chlorpromazine and haloperidol revolutionized the treatment of schizophrenia, dramatically reducing the stay of schizophrenics in mental hospitals and saving many billions of dollars in hospital care. Research in the past decade has shown that antipsychotic drugs also exert their effect on the catecholamine neurotransmitters. Carlsson had observed that antischizophrenic drugs caused an increase in the formation of catecholamines in the rat brain, and he formed the hypothesis that this was owing to the drug's ability to block dopamine receptors. Work by other investigators has confirmed and extended this hypothesis. Antipsychotic drugs do block dopamine receptors in the brain, and there is a strong associa-
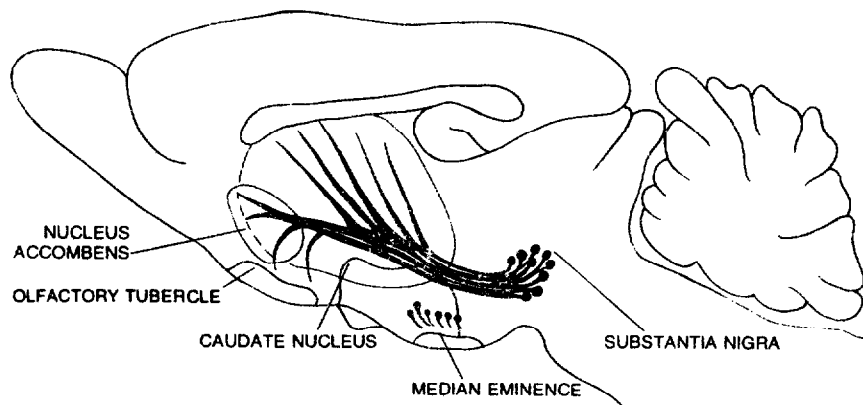


DOPAMINE TRACTS, the main bundles of nerves containing dopamine, are shown (black) in a drawing of a longitudinal section along the midline of the rat brain. The cell bodies are concentrated in the substantia nigra, and the axons project primarily to the caudate nucleus. A dopamine deficiency in that region causes Parkinsonism, which can be treated with dopa.
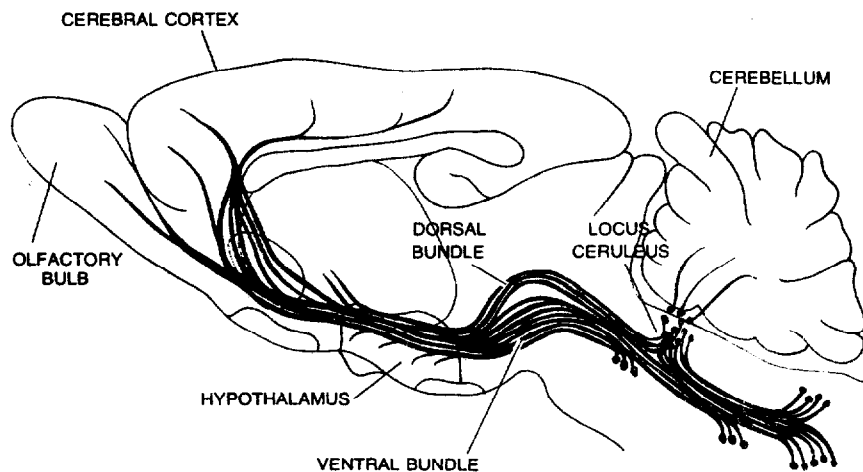


NORADRENALINE TRACTS arise primarily in the locus ceruleus and reach many brain centers, including the cerebellum, cerebral cortex and hypothalamus. Illustrations on this page are based on maps made by Urban Ungerstedt of Royal Caroline Institute in Sweden.

tion between the blocking ability of various drugs and their capacity to relieve schizophrenic symptoms. These findings point clearly to the involvement of the dopaminergic nerves in schizophrenia.

Amphetamine has also helped to clarify the nature of schizophrenia. Taken repeatedly in large amounts, amphetamine produces a psychosis manifested by repetitive and compulsive behavior and hallucinogenic delusions that are indistinguishable from the symptoms exhibited by paranoid schizophrenics. Amphetamine releases catecholamines from nerves in the brain to stimulate both noradrenergic and dopamine receptors. After doing experiments with two forms of amphetamine Solomon H. Snyder of the Johns Hopkins University Medical School hypothesized that the schizophrenia-like psychosis the drug induces is due to excessive release of dopamine.

The ability of antischizophrenia drugs, which block dopamine receptors, to relieve symptoms of amphetamine psychosis is consistent with this hypothesis.

Although there have been rapid advances in our knowledge of neurotransmitters in the past 20 years, much remains to be discovered about these compounds. Only a few of the chemical transmitters of the brain neurons have even been characterized. The role of neurotransmitters in behavior, mood, reproduction and learning and in diseases such as depression, schizophrenia, motor disorders and hypertension is beginning to evolve. If only the present trend toward reducing the funds committed to research support can be reversed, exciting new discoveries about neurotransmitters should soon be made, many of which will surely contribute directly toward the treatment or cure of some of man's most tragic afflictions.